

CLAIMS

What is claimed is:

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1) A *Brassica* plant comprising a DNA fragment including a fertility restorer locus for Ogura cytoplasmic male sterility, wherein said DNA fragment can be identified through at least one marker of bin 2, but cannot be detected by at least one marker of bin 3.

10 2) A *Brassica* plant comprising a DNA fragment including a fertility restorer locus for Ogura cytoplasmic male sterility, wherein said DNA fragment can be identified through at least one marker of bin 2, but none of the markers of bin 3.

15 3) A *Brassica* plant according to claim 2 comprising a DNA fragment including a fertility restorer locus for Ogura cytoplasmic male sterility, wherein said DNA fragment can be identified through all the markers of bin 2, but none of the markers of bin 3.

4) The *Brassica* plant according to anyone of claim 1 - 3, wherein bin 2 is comprised of the markers E33M47, E2M4-1, E3M1-1, E4M14-1, E5M1-2, E5M4-2, and E8M14-2.

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5) The *Brassica* plant according to anyone of claim 1 - 3, wherein bin 3 is comprised of the markers OPY17, OPN20, and E8M1-2.

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6) The *Brassica* plant according to claim 4, wherein said markers are amplified in a polymerase chain reaction using primer pairs represented by 1159 and 1160; E2 and M4; E3 and M1; E4 and M14; E5 and M1; E5 and M4; E8 and M14, respectively.

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7) The *Brassica* plant according to claim 5, wherein said markers are amplified in a polymerase chain reaction using the primer pairs represented by PR0004F and PR0004R; 1135 and 1136; and E8 and M1, respectively.

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8) The *Brassica* plant according to anyone of claim 1 - 7, wherein said DNA fragment is the BLR1 recombination event.

5 9) The *Brassica* plant according to anyone of claim 1 - 8, wherein said plant is an inbred plant.

10 10) The *Brassica* plant according to anyone of claim 1 - 8, wherein said plant is a hybrid plant.

10 11) The *Brassica* plant according to claim 8, wherein said BLR1 recombination event is obtainable from the *Brassica* inbred line BLR-038, a sample of the seed of inbred line BLR-038 having been deposited with NCIMB under accession number NCIMB 41193.

15 12) A method of detecting a *Brassica* plant containing a restorer gene, comprising the steps of:

- a) obtaining a sample from a *Brassica* plant;
- b) detecting in said sample a DNA fragment by
 - i) at least one marker of bin 2, but not by at least one marker of bin 3;
 - ii) at least one marker of bin 2, but none of the markers of bin 3;
 - iii) all the markers of bin 2, but none of the markers of bin 3

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13) The method of detecting a *Brassica* plant according to claim 12, further comprising selecting said *Brassica* plant, or a part thereof, containing said DNA fragment.

25 14) The method of detecting a *Brassica* plant according to claim 12, further comprising the step of selfing said *Brassica* plant containing said DNA fragment.

15) The method of detecting a *Brassica* plant according to claim 12, further comprising the step of crossing said *Brassica* plant with another *Brassica* plant.

16) The method of selecting a *Brassica* plant according to anyone of claims 12 - 15, wherein said DNA fragment comprises the BLR1 recombination event.

17) The method of selecting a *Brassica* plant according to claim 12, wherein said marker of
5 bin 2 comprises E33M47, E2M4-1, E3M1-1, E4M14-1, E5M1-2, E5M4-2, or E8M14-2.

18) The method of selecting a *Brassica* plant according to claim 12, wherein said marker of
bin 2 has partial homology to E33M47, E2M4-1, E3M1-1, E4M14-1, E5M1-2, E5M4-2,
or E8M14-2.

10 19) The method of detecting a *Brassica* plant according to claim 12, further comprising the
step of detecting in said sample a DNA fragment obtainable by PCR amplification using
primers 1159 and 1160, whereas said DNA fragment is not amplified by the primers
PR0004F and PR0004R.

15 20) A combination of markers for detecting the presence of the BLR1 recombination event,
comprising a marker of bin 2 and a marker of bin 3.

21) The combination of markers for detecting the presence of the BLR1 recombination event
20 according to claim 18, wherein said marker of bin 2 comprises the markers E33M47,
E2M4-1, E3M1-1, E4M14-1, E5M1-2, E5M4-2, or E8M14-2 and wherein said marker of
bin 3 comprises OPY17, OPN20, or E8M1, or a marker having partial homology to any
one of these markers.

25 22) A method for screening a *Brassica* plant to determine whether it contains the BLR1
recombination event, comprising extracting DNA from said *Brassica* plant, subjecting the
extraction to a polymerase chain amplification reaction in the presence of DNA fragments
represented by primers 1159, 1160, PR0004F, and PR0004R, and determining the
amplification of DNA fragments from the extracted DNA by primers 1159 and 1160 and
30 lack of amplification of DNA fragments from extracted DNA that correspond to primers
PR0004F and PR0004R.

23) A method for producing a fertile F1 hybrid *Brassica* plant comprising the steps of
5 a) determining total glucosinolate content in the male fertile restorer parent comprising
 the BLR1 recombination event and, optionally, also in the female male sterile CMS
 parent;
 b) crossing the female and male parents to produce F1 hybrid seed.

24) A method for producing a fertile F1 hybrid *Brassica* plant comprising the steps of
10 a) detecting in seed or a plant of the male fertile restorer parent the BLR1 recombination
 event through marker analysis;
 b) crossing the female and male parents to produce F1 hybrid seed.

15 25) A method according to anyone of claims 23 - 24 comprising the additional step of
 detecting in seed or a plant of the restorer parent a DNA fragment through marker
 analysis.

20 26) The method for producing a fertile F1 hybrid *Brassica* plant according to to anyone of
 claims 23 - 25, comprising the additional step of planting said F1 hybrid seed.

25 27) The method for producing a fertile F1 hybrid *Brassica* plant according to to anyone of
 claims 23 - 26, comprising the additional step of harvesting the F2 seed grown from the
 plant resulting from said F1 seed.

28) A method according to claim 27 comprising the additional step of determining total
25 glucosinolate content in F2 seed derived from the F1 hybrid plant.

29) A hybrid F1 *Brassica* plant produced by the method of to anyone of claims 23 - 28.

30 30) A *Brassica* plant comprising the BLR1 recombination event, wherein said event is
 obtainable from the *Brassica* inbred line BLR-038, a sample of the seed of inbred line
 BLR-038 having been deposited with NCIMB under accession number NCIMB 41193.

31) A method for producing a *Brassica* plant containing the BLR1 recombination event comprising the steps of obtaining a *Brassica* plant containing the BLR1 recombination event, crossing this plant with another *Brassica* plant, obtaining hybrid seed produced by this cross, and planting said hybrid seed to produce a *Brassica* plant containing the BLR1 recombination event.

5 32) A kit for detecting the BLR1 recombination event comprising:

- a) a first pair of primers that amplify a marker of bin 2; and
- 10 b) a second pair of primers that does not amplify a marker of bin 3.

33) A *Brassica* plant comprising the BLR1 recombination event.

34) The *Brassica* plant according to claim 29, wherein said BLR1 recombination event is
15 obtainable from the *Brassica* inbred line BLR-038.

35) The *Brassica* plant according to anyone of claims 1 – 11; 29, 30, 33, and 34, wherein
said plant is a *Brassica napus*, *Brassica campestris*, *Brassica oleracea*, *Brassica nigra*,
20 *Brassica carinata* or any other species belonging to the *Brassicaceae* family.

36) The *Brassica* plant according to claim 35, wherein said plant is a sexual or asexual
recombination or clone of said species.

37) The *Brassica* plant according to anyone of claims 1 – 11; 29, 30, and 33 - 35, said plant
25 comprising a total glucosinolate level equal to or lower than the glucosinolate levels of
double-low *Brassica* varieties.

38) A *Brassica* plant comprising a recombination event resulting from a break between the
fertility restorer locus for Ogura cytoplasmic male sterility derived from the Ogura
30 *Raphanus sativus* and the glucosinolate locus along a nucleic acid segment and
subsequent rejoining to produce a new nucleic acid segment, which plant exhibits fertility

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restoring activity resulting from expression of the *Raphanus sativus* restorer gene and a GSL content no higher than that normally found in double low open pollinated varieties, but preferably in a range of between 0.5 to 18 μ mol total glucosinolate (GSL) per gram (g) of seed at 9% humidity, particularly in a range of between 2 and 15 μ mol total glucosinolate (GSL) per gram (g) of seed at 9% humidity, more particularly in a range of between 3 and 14 μ mol total glucosinolate (GSL) per gram (g) of seed at 9% humidity, but especially a GSL content of between 3.5 and 10 μ mol total glucosinolate (GSL) per gram (g) of seed at 9% humidity.

10 39) A Brassica plant according to claim 38, wherein the GSL content is in a range of between 3.6 and 6.0 μ mol, but especially between 3.6 and 4.2 μ mol total glucosinolate (GSL) per gram (g) of seed at 9% humidity.